

Polyclonal Antibody to PPFA2 - Aff - Purified

Catalog No.:	15-288-21472
Quantity:	0.1 mg
Concentration:	1.0 mg/ml
Background:	Connective Tissue Growth Factor (CTGF) is an immediate early growth responsive gene that has been shown to be a downstream mediator of TGFbeta actions in fibroblasts and vascular endothelial cells. CTGF promotes cell adhesion and mitogenesis in both fibroblasts and endothelial cells and stimulates cell migration in fibroblasts. The localization of CTGF in angiogenic tissues, as well as in atherosclerotic plaques, suggests a possible role in the regulation of vessel growth during development, wound healing, and vascular disease. CTGF is expressed in myocardial infarct tissue following acute myocardial infarction.
Host / Isotype:	Chicken
Immunogen:	Recombinant human CTGF
Format:	State: Concentrated tissue culture supernatant, dialyzed, lyophilized Buffer System: PBS, pH 7.2 containing 0.09% Sodium Azide as preservative. Reconstitution: Restore with 0.5 ml distilled water.
Applications:	Immunohistochemistry on Frozen Sections (2.5-5 µg/ml). Immunohistochemistry on Paraffin Embedded Sections (20 µg/ml). Microwave pretreatment in citrate buffer is recommended for antigen retrieval. Suggested Positive Control: Human placenta, Rat spleen. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	This antibody detects CTGF. This monoclonal antibody also stains cells in rat spleen, indicating significant cross reactivity with the corresponding Rat CTGF. Species: Human and Rat. Others not tested.
Storage:	Store the original vial at 2-8°C, stock solution or aliquots thereof at -20°C. Avoid repeated freezing and thawing. Shelf life: One year from despatch.
General References:	1. Schwab J.M. et al. (2000) Connective tissue growth factor (CTGF) is expressed by a subset of reactive astrocytes. Localization and accumulation in human cerebral infarctions. Neuropathol Appl Neurobiol 26: 434-440 2. Schwab JM, Beschoner R, Nguyen TD, Mittelbronn M, Schluesener HJ : Differential cellular accumulation of connective tissue growth factor defines a subset of reactive astrocytes, invading fibroblasts, and endothelial cells following central nervous system

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injury in rats and humans. J. Neurotrauma. 2001 Apr; 18(4):377-388

Protocols:**IMMUNOHISTOCHEMISTRY WITH UNLABELED MONOCLONAL ANTIBODIES ON PARAFFIN-EMBEDDED TISSUE SECTIONS**

1. Deparaffinize slides in chloroform for 30min and rehydrate to distilled water
2. Antigen retrieval: microwave treatment (700W) for 15min in 300ml 0.01M citrate buffer pH 6.0.
3. Incubate slides for 15min in 200ml methanol/4.8ml 30%H₂O₂
4. Rinse slides 2x in distilled water and for 5min in TBS, pH 7.5.
5. Block with normal swine serum (NSS, Jackson ImmunoResearch #014-000-121) diluted 1/10 in Tris buffered saline pH 7.5, containing 0.1% bovine serum albumin (BSA, Calbiochem). Cover sections with NSS and incubate for 15min in a humid chamber at room temperature (RT).
6. Remove serum from sections (do not rinse)
7. Incubate with primary antibody diluted in TBS/0.1%BSA, pH 7.5 for 60min in a humid chamber at room temperature
8. Rinse sections in TBS, pH 7.5 for 5min, 2x
9. Incubate with secondary antibody (rabbit anti mouse IgG+IgM-biotin, Dako E 0464) 1:400 diluted in TBS/0.1%BSA, pH 7.5 for 30min in a moist chamber, room temperature. Preincubate now StreptABComplex /HRPO in TBS/0.1%BSA, pH 7.5!
10. Rinse sections in TBS, pH 7.5 for 5min, 2x
11. Incubate with StreptABComplex /HRPO (DAKO, K 0377) for 30min in a humid chamber at room temperature
12. Rinse sections in TBS, pH 7.5 for 5min, 2x
13. DAB-staining: Prepare 3,3'-Diaminobenzidintetrahydrochloride (DAB) according the manufacturers instructions (e.g. DAB tablets) and incubate slides for 1-15 min. Check color reaction to avoid overstaining.
14. Rinse sections in TBS, pH 7.5 for 5min, 2x
15. Counterstain in haematoxylin for 1min and rinse in cold tap water for 10min
16. Mount slides with coverslips.

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